THE PHOSPHORUS REQUIREMENTS OF THE CHICKEN WITH SPECIAL REFERENCE TO THE AVAILABILITY OF PHYTIN PHOSPHORUS

BY

EDWIN PIERCE SINGSEN

B. S., Rhode Island State College, 1938

M. S., North Carolina State College, 1940

ABSTRACT OF A THESIS

SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY IN ANIMAL NUTRITION IN THE GRADUATE SCHOOL OF THE UNIVERSITY OF ILLINOIS, 1945

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THE PHOSPHORUS REQUIREMENTS OF THE CHICKEN WITH SPECIAL REFERENCE TO THE AVAILABILITY OF PHYTIN PHOSPHORUS

During the first few weeks of life the growing chick has a relatively high phosphorus requirement. Bethke, Kennard, Kick and Zinzalian (1929) showed that 0.37 per cent phosphorus in the ration supported very good bone formation and satisfactory growth. Bethke, Kennard and Kick (1929) presented evidence that 0.44 per cent phosphorus supported maximum bone calcification. Hart, Scott, Kline and Halpin (1930) observed that in the presence of adequate vitamin D, 0.31 per cent phosphorus supported good growth and bone calcification. Mitchell and McClure (1937) suggest that 0.45 per cent phosphorus in the diet will meet the minimum requirements of the chick. In general, the phosphorus comes from two sources, (1) ingredients of animal origin and (2) ingredients of plant origin, the phosphorus contributed by the latter group of ingredients usually being considered the least available of the two. Common (1940) reported that 50 to 75 per cent of the total phosphorus in many poultry feed ingredients of plant origin is in the form of phytin, the calcium-magnesium salt of inositol hexaphosphoric acid. According to Lowe, Steenbock and Krieger (1939), Common (1939) and others, the phosphorus of phytin and of cereal grains is poorly available to the growing chick and rat. Thus it appeared that (1) any quantitative measurement of the phosphorus requirement of the growing chick should be made with little or no phytin phosphorus in the diet and (2) any factors that might enhance the utilization of phytin phosphorus would be of great practical importance.

Basal diet P-4, designed to be adequate in all nutrients except phosphorus and calcium had the following percentage composition: special white corn meal (degermed and debraned) 70.5, casein 12.0, gelatin 3.0, primary grown yeast 4.0, bagasse 4.0, liver meal 3.0, potassium chloride 0.5, salt 0.5, wheat germ oil 0.25, choline chloride 0.1 and 70 grams of manganous sulphate per 100 pounds of ration. To each 100 grams of diet was added the following: 300 gamma of riboflavin, 500 gamma of pyridoxine, 1400 gamma of calcium-pantothenate, and 100

A.O.A.C. chick units of vitamin D. A calcium: phosphorus ratio of approximately 2:1 was used in all experiments.

Methods

The feed intakes of the lots in each experiment were equalized on a grams per chick per day basis, the amount actually fed being set by the lot consuming the least. Total calcium and phosphorus were determined by the A.O.A.C. (1940) volumetric procedures. The tibia were ashed individually and the results expressed on the basis of the dry, fat-free, bone. The methods used to determine phytin phosphorus and phytase activity have been summarized briefly in the following paragraphs.

Phytin phosphorus was determined by a modification of the method of Common (1940). Approximately 2 grams of feed was extracted with half-normal hydrochloric acid, the extract filtered, neutralized to phenolphthalein with sodium hydroxide and heated on a water bath after the further addition of a dilute solution of ferric chloride. The precipitate was removed by centrifuging, washed with half-normal acid and recentrifuged. The precipitate was then heated after the addition of a two per cent solution of sodium hydroxide, filtered, the filtrate ashed and total phosphorus determined on the residue.

Phytase activity was determined by bringing together five ml. of an aqueous extract of the feed being examined and five ml. of a sodium phytate substrate. The reaction mixture was adjusted to pH 5.2 and after six hours the reaction was stopped with tri-chlor-acetic acid and inorganic phosphorus determined. The enzyme activity was expressed as the per cent of substrate phosphorus liberated.

Results

Two series of experiments were conducted to determine (1) the total inorganic phosphorus requirements of the chicken and (2) the level of phytin phosphorus needed in the diet to meet these requirements. Sodium acid phosphate and commercial calcium-magnesium phytate were used to supplement the low-phosphorus basal diet such that when the diet contained 0.45 per cent total phosphorus the supplement contributed over 50 per cent of it. The quantitative inorganic phosphorus requirement for satisfactory growth was 0.340 per cent of the diet, or less, while that for satisfactory bone calcification was between 0.386 and 0.471 per cent. These results are in good agreement

with those of Bethke, Kennard and Kick (1929). The data clearly indicate that the phosphorus requirement for growth is less than the requirement for good bone calcification. Commercial calcium-magnesium phytate supported good growth but failed to support satisfactory bone calcification.

Among the factors that appear to condition the utilization of phytin phosphorus are (1) the plant enzyme phytase and (2) vitamin D. A preliminary experiment indicated that the introduction of field-cured alfalfa leaf meal, supposedly high in phytase activity, into a soybean meal chick diet, enhanced the utilization of the prosphorus in the diet. When, however, phytase was extracted from wheat bran and added to the low phosphorus diet supplemented with commercial calcium-magnesium phytate, it failed to enhance the utilization of the phytin phosphorus. When wheat bran, containing phytin phosphorus and active phytase, was added to the low-phosphorus diet, the enzyme failed to enhance the utilization of the phosphorus present in the bran.

The vitamins D of irradiated animal sterols and cod-liver oil were compared at several levels of A.O.A.C. unitage for their ability to promote the utilization of phytin phosphorus for bone calcification. At all levels fed the irradiated animal sterols promoted better calcification, the average ratio of efficiency being 1:1.34. Increasing amounts of vitamin D from both sources also improved bone calicification. McGinnis, Norris and Heuser (1944) have also observed the correlation between the level of vitamin D and the utilization of phytin phosphorus.

Conclusions

On the basis of the data obtained, subject to the experimental conditions and the strain of chicks used, the following conclusions seem justified:

1. The quantitative requirement of the chick for total inorganic phosphorus for growth is less than its requirement for satisfactory bone calcification.

2. The quantitative requirement for growth appears to be satisfied by 0.340 per cent, or less, of total inorganic phosphorus in the diet.

3. The quantitative inorganic phosphorus requirement for satisfactory bone calicification appears to lie between 0.386 and 0.471 per cent for the four-week old chick, and between 0.394 and 0.443 per cent for the eight-week old chick.

4. Commercial calcium-magnesium phytate is not a satisfactory phosphorus supplement in the diet of the growing chick.

5. The phosphorus of phytin can apparently support growth by functioning in certain metabolic processes necessary for growth, without being split off from the molecule as inorganic phosphate.

6. The phosphorus of the intact phytin molecule does not appear to be deposited in the mineral salts formed during bone calcification.

7. The presence of active phytase in the diet does not enhance the utilization of the phosphorus of commercial calcium magnesium phytate by the growing chick under the conditions of these experiments.

8. No evidence was obtained that active phytase, contributed to the diet by wheat bran, enhances to any appreciable extent the utilization of the prosphorus of wheat bran by the growing chick.

9. The field-cured alfalfa leaf meal used in experiment 1 definitely enhanced the utilization of the plant phosphorus present in diet P-1.

10. There is a correlation, curvilinear in nature, between the level of vitamin D fed and the average per cent bone ash produced by chicks receiving phytin as their chief source of dietary phosphorus.

11. The vitamin D found in irradiated animal sterols is more effective on an equal A.O.A.C. unitage basis in enhancing the utilization of phytin phosphorus by the growing chick than is the vitamin D of cod-liver oil.

12. Although increasing amounts of vitamin D enhance the utilization of phytin phosphorus by the growing chick, the bone calcification produced never equals that obtained when the diet is supplemented with inorganic phosphorus.

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VITA

The author was born at Hackensack, New Jersey, on December 10, 1916. He received the B. S. degree from the Rhode Island State College in 1938 and the M. S. degree from North Carolina State College in 1940, receiving the Phi Kappa Phi scholarship award for the graduate students from the latter institution in 1940. He served as research assistant in poultry husbandry at the North Carolina State College from 1938 to 1940, as instructor in poultry husbandry at the University of Connecticut 1940 to 1941, and as an assistant in poultry husbandry at the University of Illinois from 1941 to 1943. In 1943 he returned to the University of Connecticut as assistant professor of poultry husbandry; the position which he holds at the present time. The author is a member of Alpha Zeta.

A list of publications follows:

Singsen, E. P. 1940. The Pathological Histology of Certain Avian Neoplasms. Master Thesis. North Carolina State College.

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